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High Male:Female Ratio of Germ-Line Mutations: An Alternative Explanation for Postulated Gestational Lethality in Males in X-Linked Dominant Disorders

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Summary

In this paper I suggest that a vastly higher rate of de novo mutations in males than in females would explain some, if not most, X-linked dominant disorders associated with a low incidence of affected males. It is the inclusion of the impact of a high ratio of male:female de novo germ-line mutations that makes this model new and unique. Specifically, it is concluded that, if an X-linked disorder results in a dominant phenotype with a significant reproductive disadvantage (genetic lethality), affected females will, in virtually all cases, arise from de novo germ-line mutations inherited from their fathers rather than from their mothers. Under this hypothesis, the absence of affected males is explained by the simple fact that sons do not inherit their X chromosome (normal or abnormal) from their fathers. Because females who are heterozygous for a dominant disorder will be clinically affected and will, in most cases, either be infertile or lack reproductive opportunities, the mutant gene will not be transmitted by them to the next generation (i.e., it will be a genetic lethal). This, not gestational lethality in males, may explain the absence of affected males in most, if not all, of the 13 known X-linked dominant diseases characterized by high ratios of affected female to male individuals. Evidence suggesting that this mechanism could explain the findings in the Rett syndrome is reviewed in detail.

Introduction

As discussed in detail by James Crow (1993), Drost and Lee (1995), and others (see discussion below), there is a great deal of evidence indicating that the mutation rate is much higher in human males than in females. Findings supporting this conclusion include the greater rate of evolution of Y pseudogenes than those on the autosomes, increased paternal age associated with new dominant mutations, and the increase paternal:maternal contribution to de novo X-linked recessive disorders. These data, combined with other evidence, have given rise to the concept of "male-driven molecular evolution." (Miyata at al. 1990; Shimmin et al. 1993).

Evidence that this phenomenon may also have direct clinical relevance is presented in table 1, which lists 13 disorders associated with an excess of affected female to male patients. In most of these disorders, the discrepancy in the numbers of affected males and females has been attributed to gestational lethality in males. For a review of the evidence for this mode of inheritance, see Wettke-Schäfer and Kantner (1983). It is, however, difficult to determine from the current literature whether this is the correct explanation for the deficiency of affected males in these disorders. To date, the evidence is based on a small number of case reports, often without evidence for, or statistical analysis of, an increase in the frequency of spontaneous abortions. Also, for some of these disorders, an occasional affected male is described.

Thus, although these disorders are generally considered to be examples of X-linked dominant abnormalities with male lethality, there is little direct evidence to support this conclusion. It, therefore, appears reasonable to test other hypotheses that might explain the deficiency of affected males in these disorders. For reasons outlined below, a high ratio of male to female mutations is a reasonable alternative explanation.

Evidence for High Male:Female Mutation Rate

Evidence for a much higher rate of de novo mutations in males than in females was suggested almost 50 years ago. In 1947, Haldane noted that certain types of de novo genetic alterations occur more frequently in males than in females. Although the evidence supporting this hypothesis was sparse initially, there is now considerable direct evidence that it is, in fact, correct. For reviews

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Table 1

X-Linked Dominant Inherited Diseases, Characterized by High Ratios of Female to Male Patients, Currently Attributed to or Suggested to Result from Gestational Lethality in Hemizygous Males

Disorder	McKusick Catalog No.[a]
Incontinentia pigmenti (Bloch-Sulzberger syndrome)	308310
Oral-facial-digital type 1 (OFD1 syndrome)	311200
Focal dermal hypoplasia (Goltz syndrome)	305600
Chondrodysplasia punctata, X-linked dominant (Conradi-Hunermann or Happle syndrome)	302960
Cataract, congenital, with microcornea or slight microphthalmia	302300
Lipodystrophy, familial partial (Kobberling-Dunnigan syndrome)	308980
Corpus callosum, agenesis of, with chorioretinal abnormality (Aicardi syndrome)	304050
Autism, dementia, ataxia, and loss of purposeful hand use (Rett syndrome)	312750
Ichthyosiform erythroderma, unilateral, with ipsilateral malformations (CHILD syndrome)	308050
Microphthalmia with linear skin defects (MLS) (Midas syndrome)	309801
Serpentine fibula-polycystic kidney syndrome (SFPKS)	600330
Cerebral, ocular, dental, auricular and skeletal abnormalities (CODAS syndrome)	600373
Cervico-oculo-acoustic (Wildervanck syndrome)	314600

[a] McKusick (1994) with additions from OMIM™, December, 1995.

and interpretations of the evidence for differences in mutation rates between males and females, see Vogel (1977), Vogel and Motulsky (1986), Crow (1993), and Shimmin et al. (1993). Also, for a detailed discussion of the biological basis and the rates of spontaneous germline mutations, see Drost and Lee (1995). By use of molecular markers, direct evidence for a sexual bias in the origin of mutations has been shown for ornithine transcarbamylase (OTC) deficiency (Bonaiti-Pellié et al. 1990; Tuchman et al. 1995); hemophilia A (Bröcker-Vriends et al. 1991; Rossiter et al. 1994); hemophilia B (Kling et al. 1992; Ketterling et al. 1993), and the Lesch-Nyhan syndrome (Francke et al. 1976, 1977; Vogel 1977).

In a study of 43 hemophilia B families, it was found that, while the male:female ratio of all point mutations was 3.5:1, the ratio of transitions at CpG dinucleotides was 11:1 (Ketterling et al. 1993). This supports, at least in part, a model involving 5-methyl cytosine deamination (Barker et al. 1984; Cooper and Youssoufian 1988; Koeberl et al. 1990). Tuchman et al. (1995) in a study of OTC families found that, whereas 12/15 (80%) of heterozygous females resulted from de novo mutations, only 2/28 (7%) of the mutations in affected males were sporadic. From these data, they concluded that the "... mutation rate in the OTC gene in male germ cells is about 50-fold higher than in female germ cells" (Tuchman et al. 1995, p. 68). While there were insufficient data to permit an estimate of the rates of specific mutation types, their data also suggest an excess of CpG dinucleotide mutations.

In contrast, other disorders with a high male:female mutation rate result not from single base changes but from gross chromosomal abnormalities. This, for example, is what has been found in the de novo mutations in ~45% of a severe form of hemophilia A (Rossiter et al. 1994). In families with this disorder in which the chromosomal abnormality originated in a maternal grandparent, DNA haplotype analysis indicated that 20/20 alterations occurred in the germ line of the grandfather. In addition, 49/50 mothers of sporadic male cases of this form of hemophilia A were carriers (Rossiter et al. 1994). Earlier work had demonstrated that these cases of hemophilia A resulted from intrachromosomal inversions of the region between gene A within intron 22 of the factor VIII gene and a copy of gene A located telomerically to the factor VIII gene (Lakich et al. 1993; Naylor et al. 1993). Thus de novo inversions responsible for ~45% of severe hemophilia A cases occur in male germ cells with an estimated male:female ratio of $\geq$302:1 (Rossiter et al. 1994).

A striking alteration in the sex ratio of the mutation rate has also been found in Charcot-Marie-Tooth type 1A (CMT1A) disease. Palau et al. (1993) found that sporadic cases resulting from de novo duplications arose from unequal crossing-over between chromosome 17 homologues during paternal meiosis. As with the chromosomal abnormality found in hemophilia A, the average paternal age did not differ from that of the control group. The lack of paternal age effect in these latter disorders is in keeping with the suggestion that chromosomal abnormalities of this type arise during the one male meiosis per gamete.

Predictions of the Model

On the basis of these findings and supporting data, I conclude that marked elevations in the rate of de novo mutations in males as compared to females is the rule, not the exception. In addition, direct molecular studies indicate that this difference in mutation rates in the germ cells of males and females also occurs in genes located on the X chromosome. Moreover, when paternal, X-linked mutations have as their consequence reduced fertility of affected females (i.e., X-linked dominant disorders severe enough to prevent reproduction in affected female offspring), several predictions can be made:

- Almost all cases (except MZ twins) would be sporadic.
- Affected individuals would be exclusively females.
- Affected families would show no increase in male lethality.
- Affected families would show no increase in spontaneous abortions.
- Almost all DZ twins would be discordant.
- MZ twins would be concordant.
- Mothers and fathers of the patients would be noncarriers.
- An affected female would have a 50% chance of having an affected (male or female) offspring in the unlikely event of a pregnancy.
- The de novo mutations would be found on the paternally derived X chromosome.

Rett Syndrome (RS) as a Paradigm

Because population and family data are more extensive for RS than for other disorders listed in table 1, I have used this disorder as a model to examine these predictions. RS is characterized by a constellation of clinical abnormalities including progressive encephalopathy, severe mental retardation, and stereotypic hand movements (Trevathan and Naidu 1988; Hagberg et al. 1993). With rare exceptions, RS occurs only as isolated (nonfamilial) cases in females.

That RS has a genetic etiology is supported by the observation that MZ twins are concordant and DZ twins are disconcordant for the disorder (Naidu et al. 1988; Zoghbi 1988). The fact that, excluding MZ twins, virtually all cases of RS are nonfamilial is, however, difficult to explain by the usual rules of genetic inheritance. In spite of the lack of supporting evidence, it is generally considered that RS is an X-linked abnormality with male lethality.

Hagberg et al. (1983) first suggested that RS results from a dominant mutation of an X-linked gene. It was proposed that the abnormal sex ratio of RS was the result of early deaths of male fetuses. There is, however, no evidence for either excessive fetal loss or a deficiency of males in RS families as would be expected if this explanation were correct (Migeon et al. 1995).

Comings (1986), who also invoked an X-linked dominant mutation as the cause of RS, indicated that the apparent absence of male lethality could be explained if every RS patient resulted from a de novo mutation. This, he suggested, would make it unlikely that both a RS female and evidence of male lethality would be encountered in the same family. Still left unanswered, however, is why a mutation that causes RS in females would be lethal in males. After a review of the current data, Migeon et al. (1995), however, concluded that there was no compelling evidence supporting any of the mechanisms proposed, to date, to explain the sex-limited expression of RS.

In the current proposal the exclusive occurrence of RS in females, without evidence of male lethality, is explained by the fact that de novo, X-linked mutations occurring exclusively in male germ cells could only be passed on to, and result in, affected daughters. Thus, it is the high male:female de novo germ-line mutation rate, not male lethality, that is proposed here to explain the absence of affected males in this disease.

Not only would de novo, X-linked dominant mutations in male germ cells explain the exclusive occurrence of RS in females without increased fetal wastage, it would also account for the sporadic (nonfamilial) incidence of this disorder. As pointed out by Haldane (1935, 1947), when an X-linked dominant mutation results in a reproductive lethal disorder in males without affecting the female reproductive rate, one-third of the cases would represent new mutations (Vogel and Rathenberg 1975). If, however, the mutation were a reproductive lethal disorder in both sexes, every case would represent a new mutation.

The report by Witt-Engerström and Forslund (1992) of RS in both a mother and her daughter is also compatible with the prediction that if on a rare occasion an affected female had an offspring there would be a 50% chance of it being either an affected male or female. If mutations of the types described above should occur occasionally as a germ-line mosaic in either sex, one would also expect to find rare exceptions to the above predictions. Indeed, there are a small number of familial cases of RS that cannot be explained by this proposal. Nevertheless, the fact that $\leq 1\%-2\%$ of all cases (some with atypical phenotypes) are familial supports the concept that RS, if genetic, must result in almost all cases from de novo mutations.

Future Investigation

The finding that the paternal age in RS is not increased (Murphy et al. 1986; Akesson et al. 1992) is taken as indirect evidence that the underlying genetic abnormality in this disorder might involve chromosomal alterations (not single-base changes) arising at the male meiosis. This suggestion is based on the lack of a paternal age effect in severe hemophilia A and CMT1A, both now known to be caused by chromosomal abnormalities (see previous discussion). Additional evidence supporting this notion is found in the observation that severe hemophilia A and CMT1A also exhibit the most extreme sexual bias in the origin of the alteration (i.e., arise only in male germ cells) (Palau et al. 1993; Rossiter et al. 1994).

I also conclude from both the general data and from the specific model of RS that one should consider the hypothesis that a high male:female mutation ratio could be the explanation for any X-linked dominant disorder that occurs in females in the absence of affected males or well-documented excessive fetal wastage. This would include most, if not all, of the 13 disorders listed in table 1.

Because this phenomenon would result in sporadic cases of affected females, it is likely that most disorders of this type would not be recognized as "genetic." It is, therefore, suggested that one should be alert for disorders that might be explained by this mode of inheritance.

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